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ON THE CHEMOTAXIS OF THE EARTHWORM TO CARBON DIOXIDE

By

KAGEHIDE SHIRAISHI
(白 石 景 秀)
Biological Institute, Tôhoku Uuiversity, Sendai, Japan

(With 4 figures)

Many investigators have observed that earthworms creep out and die on the surface of the ground after a heavy rain. In this regard, Darwin (1881) and other investigators advanced several explanations (Friend 1921 ; Lankester 1921 ; Merker 1926 and 1930 ; Merker and Bräunig 1927 ; Focke 1930 ; Hatai and Kim 1931 ; Nishida 1951). In particular, Hatai and Kim explained that this phenomenon is induced by accumulation of carbon dioxide in the soil of the habitat of earthworms. Recently Nishida investigated this occurrence by field observations and experiments. He ascertained by his experiments that the carbon dioxide was effective to drive the earthworm out of the ground to the surface, and suggested that the factor of the creeping out of the earthworm is the accumulation of carbon dioxide and the oxygen lack in natural habitat. But the mechanism of such phenomenon has never been made clear.

The present study was designed to make clear the chemotaxis of the earthworm to gaseous carbon dioxide, as an inquiry to solve the relation between the creeping out and the accumulation of carbon dioxide in the soil. Thus, in this study, the carbon dioxide was used as a stimulant in its gaseous state.

Before going further, the author cordially thanks Professor S. Nomura, under whose direction this work was carried out, for useful suggestions and encouragement.

MATERIAL AND METHOD

The material used in this experiment was *Eisenia foetida* (Sav.) collected near the laboratory. After the collection, the material was kept a few days in the thermostat regulated at 18°C.

The apparatus for observation is shown in figure 1 ; this is a modification of the apparatus of Nagano (1935), that was used to study the reaction of earthworms

to carbon dioxide. Before the experiment, air or carbon dioxide mixture was stored in A or B respectively. Then the dead space of *ah* or *dh* was washed with air, carbon dioxide, or gas mixture, stored in A or B. After above procedure, a worm was put into tube E, and the air in A was sent to E until the

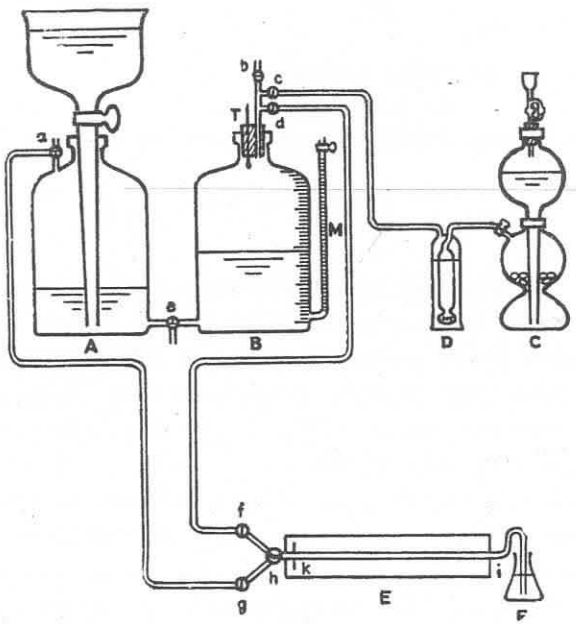


Fig. 1. Diagram of the apparatus for experiment. Detailed explanation in the text. A : Air tank, B : CO_2 mixture tank, C : Kipp's apparatus as CO_2-generator, D : Washing bottle (saturated $NaHCO_3$ solution), E : Reaction tube, F : Vent flask for exhaust gas, M : Manometer. T : Thermometer.

worm reached the mark *k*, then *h*-cock was turned open to *f* and the response of the worm was observed. At this time the velocity of the current of gas was controlled by *f*- or *g*-cocks. E was a glass tube, about 6 mm in diameter and covered with a wooden box to avoid external light that the worm might respond to.

RESULTS OF EXPERIMENT

When carbon dioxide gas reached the earthworms, they showed responses which could be classified into five groups. They are explained in table 1. The percentage of reaction of worms which reacted to various concentrations of carbon dioxide, was shown in figures 2 and 3.

The above results show that when dense carbon dioxide gas current was descending, all the worms responded to avoid carbon dioxide gas by back

or turning reaction. When the concentration of carbon dioxide was lower, the avoiding reaction diminished gradually. In general, the back reaction is the most quick avoiding motion when the strong stimulus was given to the head region of the worm ; it could also be observed when many other chemical or mechanical

Table 1

The classification of responses

Reaction	Abbreviation	Remark
Back reaction	B	The worm moves backwards, the tail takes the lead, and rhythmic movement starts from the tail.
Turning reaction	T	The worm moves round and changes the direction of progress by 180°.
Shock reaction	S	The worm stops for a moment, then progresses, not changing the direction of motion.
Accelerating reaction	A	Rhythmic movement is accelerated and the speed of progress increases.
Non-reaction	N	Remarkable change is not observed in the movements.

stimuli were given to the earthworm. If the stimuli were slightly weaker, the worm showed the turning reaction, and avoided them. To the carbon dioxide gas diluted twofold with air, the shock reaction, that the worm moves forward without change of its orientation, was often observed. It was known by the other experiment that the same proportion of carbon dioxide is very harmful to the earthworm on its physiological condition, but the worm did not avoid it. Thus the worm suffered the damage by carbon dioxide and was paralyzed at last, but the worm did not perceive it as the condition to be avoided. Next, when the gas current was ascending, the accelerating reaction appeared more frequently than any other reactions, and the rates of the back and the turning reactions were low.

Accordingly, these results may be taken as to indicate that the earthworm can perceive the direction of gas current. It would be proved more clearly, if the gradient of concentration of gas could be controlled at will. But at least, the present experiment shows that the earthworm responds differently, as the direction of current of carbon dioxide gas is ascending or descending. And the direction of movement after response, if any, was as to avoid carbon dioxide.

The above results reveal the presence in the earthworm of negative chemotaxis to carbon dioxide gas. Furthermore, experiments were carried out to inquire the reaction of the decerebrate worm to pure carbon dioxide, by the similar method as above. The results are shown in figure 4, compared with the results of the brain-intact worm. It will be seen from the figure that the decerebrate worm

shows various responses, contrary to the brain-intact worm showing the tendency to avoid carbon dioxide. In this regard, it seems probable that the decerebrate worm can not decide their orientation without the coordination as in a whole intact body, that is controlled by the brain, even if the gradient of concentration of

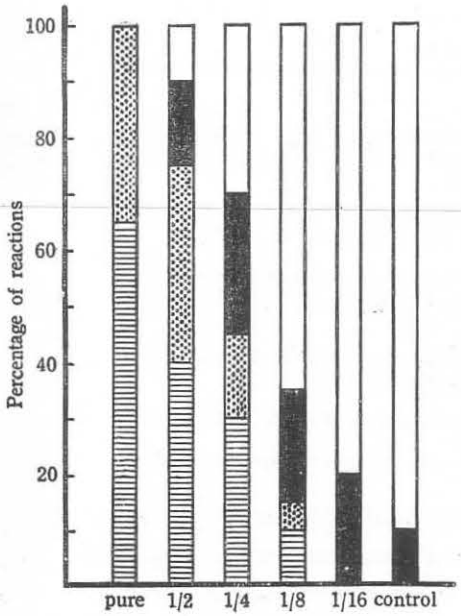


Fig. 2.

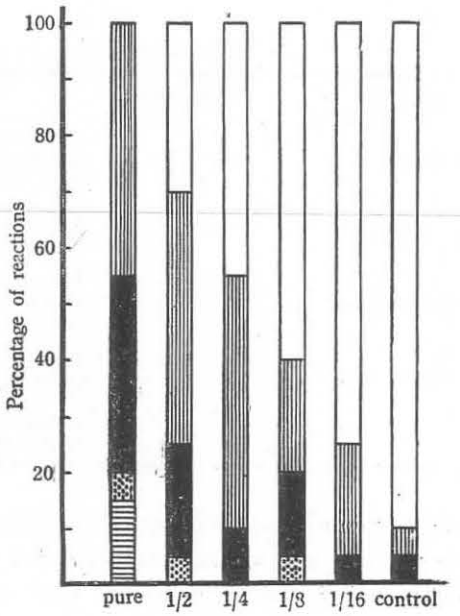
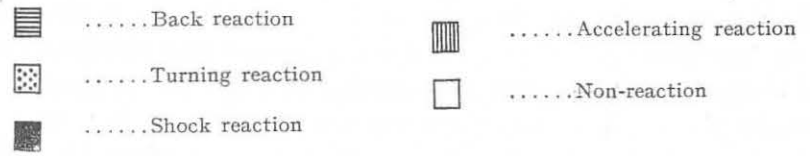


Fig. 3.

Fig. 2. The pattern of reaction to various concentrations of carbon dioxide. Ordinate, percentage of occurrence of each reaction. The direction of gas current is descending (Antero-posterior).

Fig. 3. Same as above, but the direction of gas current is ascending (Postero-anterior).



carbon dioxide were perceived by their receptors.

From these results, it may be concluded generally, that the earthworm perceives carbon dioxide as a stimulant, and shows the tendency to avoid it. But thc percentage of the non-reacting worm to carbon dioxide, diluted with air above eightfold, is considerably high. However, it has been known that the carbon dioxide in that proportion is not innocuous for the earthworm. From the other

experiment, it was found that the earthworm was paralyzed in its movement by carbon dioxide, diluted with air sixteenfold at maximum. The previous investigators suggested, that the increase of the tension of carbon dioxide in the soil may be a factor of the phenomenon that many earthworms creep out to the surface of

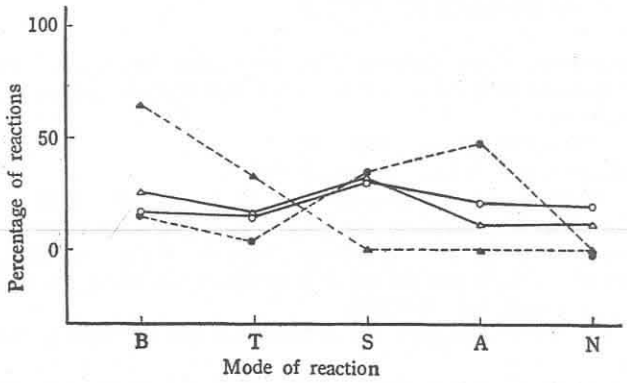


Fig. 4. The comparison of the brain-intact and the decerebrate worms.

	Brain-intact	Decerebrate
Descending	▲	△
Ascending	●	○

the ground after a heavy rain. In this respect, the results of the present experiment may support above suggestion, but it may be doubtful that the negative chemotaxis of the earthworm to carbon dioxide is caused by the change of carbon dioxide tension in the soil, which is only several per cent of the soil gas. Consequently it seems uncertain to explain the creeping out of the earthworm on the rainy day only by its chemotaxis to carbon dioxide. And further studies are necessary to resolve this problem.

SUMMARY

1. The experiment on the chemotaxis of the earthworm *Eisenia foetida* (Sav.) to carbon dioxide was carried out.

2. The earthworm showed the responses which could be classified into five groups : back reaction, turning reaction, shock reaction, accelerating reaction, and non-reaction.

3. When dense carbon dioxide was streamed descendingly to the earthworms, most of them showed the back or the turning reaction, and the accelerating reaction appeared at a high percentage to the ascending carbon dioxide current. Accordingly, it may be said that the earthworm perceives dense carbon dioxide as an unfavourable stimulant and avoid it.

4. If the brain was removed, the worm showed various responses to carbon dioxide, contrary to the brain-intact worm which showed the tendency to avoid

carbon dioxide.

5. It is difficult to explain the phenomenon, that the earthworm creeps out of the ground on a rainy day, only by its negative chemotaxis to carbon dioxide.

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